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Sequence Listing could not be accepted.

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217-9197 (toll free).

Reviewer: markspencer

Timestamp: Mon Jul 23 15:47:18 EDT 2007

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Reviewer Comments:

For SEQ ID # 16, n at location 14 can only represent a single
nucleotide.

Application No: 10523729

Version No: 1.0

Input Set:

Output Set:

Started: 2007-07-19 15:06:15.684

Finished: 2007-07-19 15:06:16.825

Elapsed: 0 hr(s) 0 min(s) 1 sec(s) 141 ms

Total Warnings: 35

Total Errors: 0

No. of SeqIDs Defined: 51

Actual SeqID Count: 51

Error code	Error Description
W 213	Artificial or Unknown found in <213> in SEQ ID (17)
W 213	Artificial or Unknown found in <213> in SEQ ID (18)
W 213	Artificial or Unknown found in <213> in SEQ ID (19)
W 213	Artificial or Unknown found in <213> in SEQ ID (20)
W 213	Artificial or Unknown found in <213> in SEQ ID (21)
W 213	Artificial or Unknown found in <213> in SEQ ID (22)
W 213	Artificial or Unknown found in <213> in SEQ ID (23)
W 213	Artificial or Unknown found in <213> in SEQ ID (24)
W 213	Artificial or Unknown found in <213> in SEQ ID (25)
W 213	Artificial or Unknown found in <213> in SEQ ID (26)
W 213	Artificial or Unknown found in <213> in SEQ ID (27)
W 213	Artificial or Unknown found in <213> in SEQ ID (28)
W 213	Artificial or Unknown found in <213> in SEQ ID (29)
W 213	Artificial or Unknown found in <213> in SEQ ID (30)
W 213	Artificial or Unknown found in <213> in SEQ ID (31)
W 213	Artificial or Unknown found in <213> in SEQ ID (32)
W 213	Artificial or Unknown found in <213> in SEQ ID (33)
W 213	Artificial or Unknown found in <213> in SEQ ID (34)
W 213	Artificial or Unknown found in <213> in SEQ ID (35)
W 213	Artificial or Unknown found in <213> in SEQ ID (36)

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Error code

Error Description

This error has occurred more than 20 times, will not be displayed

<110> Hamoen, Leendert W
Kuipers, Oscar Paul
Lindner, Johanna Cordula

<120> Novel Bacterial Expression System

<130> I-2002.012 US

<140> 10523729

<141> 2007-07-19

<150> 10/523,729

<151> 2003-08-01

<150> PCT/EP2003/008506

<151> 2003-08-01

<150> EP 02078248.8

<151> 2002-08-07

<160> 51

<170> PatentIn version 3.3

<210> 1

<211> 29

<212> DNA

<213> Bacillus subtilis

<400> 1

ttgtaaaggg acaagagctt tggataat

29

<210> 2

<211> 94

<212> DNA

<213> Bacillus subtilis

<400> 2

ttgtaaaggg acaagagctt tggataata taaaattgtg agtaatagaa ttattgctcc

60

ttgcccatta tgggcccgtt agtccaaaag gagg

94

<210> 3

<211> 113

<212> DNA

<213> Bacillus subtilis

<400> 3

gatgcagttg taaagggaca agagcttttg tataatataa aattgtgagt aatagaatta

60

ttgctccttg cccattatgg gccgcttagt ccaaaggag gtgcaaacag atg

113

<210> 4
<211> 20
<212> DNA
<213> Bacillus subtilis

<220>
<221> misc_feature
<222> (8)..(8)
<223> n is a, t, g or c

<220>
<221> misc_feature
<222> (13)..(14)
<223> n is a, t, g or c

<220>
<221> misc_feature
<222> (17)..(18)
<223> n is a, t, g or c

<220>
<221> misc_feature
<222> (20)..(20)
<223> n is a, t, g or c

<400> 4
atttaatatnta tannatnnan

20

<210> 5
<211> 33
<212> DNA
<213> Bacillus subtilis

<220>
<221> misc_feature
<222> (6)..(6)
<223> n is a, g, t or c

<220>
<221> misc_feature
<222> (11)..(13)
<223> n is a, g, t or c

<220>
<221> misc_feature
<222> (21)..(21)
<223> n is a, g, t or c

<220>
<221> misc_feature
<222> (26)..(27)
<223> n is a, g, t or c

<220>

<221> misc_feature
 <222> (30)..(31)
 <223> n is a, g, t or c

 <220>
 <221> misc_feature
 <222> (33)..(33)
 <223> n is a, g, t or c

 <400> 5
 ttattnattt nnnatttaat ntatannatn nan 33

 <210> 6
 <211> 33
 <212> DNA
 <213> Bacillus subtilis

 <400> 6
 ttattcattt ccgatttaat gtataggatg cag 33

 <210> 7
 <211> 43
 <212> DNA
 <213> Bacillus subtilis

 <400> 7
 aaaagtacat atttcttcaa aggaaaaaag caaaagatgt ttt 43

 <210> 8
 <211> 154
 <212> DNA
 <213> Bacillus subtilis

 <400> 8
 aaaagtacat atttcttcaa aggaaaaaag caaaagatgt ttttagctga aggaaaaatg 60

 aaaacgaaag ataaaaacag aggctgaaag ccatttttaa gcgtttttct tttcttggtg 120

 catcatttac aatacataca accgcaagga gagg 154

 <210> 9
 <211> 166
 <212> DNA
 <213> Bacillus subtilis

 <400> 9
 aaaagtacat atttcttcaa aggaaaaaag caaaagatgt ttttagctga aggaaaaatg 60

 aaaacgaaag ataaaaacag aggctgaaag ccatttttaa gcgtttttct tttcttggtg 120

 catcatttac aatacataca accgcaagga gaggaggaat cgcattg 166

<210> 10
<211> 10
<212> DNA
<213> Bacillus subtilis

<400> 10
ggtataatat 10

<210> 11
<211> 10
<212> DNA
<213> Bacillus subtilis

<400> 11
agttgtaaaag 10

<210> 12
<211> 13
<212> DNA
<213> Bacillus subtilis

<220>
<221> misc_feature
<222> (5)..(9)
<223> n is a, g, t or c

<400> 12
aaaannnnnt ttt 13

<210> 13
<211> 13
<212> DNA
<213> Bacillus subtilis

<220>
<221> misc_feature
<222> (5)..(9)
<223> n is a, g, t or c

<400> 13
ataannnnnt ttt 13

<210> 14
<211> 13
<212> DNA
<213> Bacillus subtilis

<220>
<221> misc_feature
<222> (5)..(9)

<223> n is a, t, g or c

<400> 14

aatannnnnt att

13

<210> 15

<211> 13

<212> DNA

<213> Bacillus subtilis

<220>

<221> misc_feature

<222> (5)..(9)

<223> n is a, g, t or c

<400> 15

aaaannnnnt att

13

<210> 16

<211> 27

<212> DNA

<213> Bacillus subtilis

<220>

<221> misc_feature

<222> (5)..(9)

<223> n is a, g, t or c

<220>

<221> misc_feature

<222> (14)..(14)

<223> n represents between 10 and 150 nucleotides that can be a, g, t
or c

<220>

<221> misc_feature

<222> (19)..(23)

<223> n is a, g, t or c

<400> 16

aaaannnnnt tttnaaaann nnntttt

27

<210> 17

<211> 30

<212> DNA

<213> unknown

<220>

<223> forward primer to amplify the ssb promoter region

<400> 17

gcgaagcttc caaacattga cgaagagtct

30

<210> 18
<211> 31
<212> DNA
<213> unknown

<220>

<223> Reverse primer to amplify the ssb promoter region

<400> 18
gctggatcct cggttaagca taagaaagac c 31

<210> 19
<211> 315
<212> DNA
<213> unknown

<220>

<223> PCR amplification product using Bacillus subtilis as template
source and SEQ ID NO: 17 and SEQ ID NO:18 as primers

<400> 19
gcttcctaaac attgacgaag agtctaaaaa agcagttatc gagcgtttca acaacgttct 60
gacttctaac ggtgcggaga tcaactggaac aaaggattgg ggtaaacgtc gtcttgctta 120
cgaaatcaac gatttccgcg acggtttcta ccaaatcgta aacgttcaat ctgacgctgc 180
ggcagttcaa gaatttgacc gtctagctaa gatcagtgac gatatcattc gccacattgt 240
tgttaaagaa gaagaataag caattttgaa atatataatg gtaaaagggtg gtcttttctta 300
tgcttaaccg aggat 315

<210> 20
<211> 28
<212> DNA
<213> unknown

<220>

<223> Forward primer to amplify the rpsF promoter region

<400> 20
gcgaagcttg tgactttgag cggggcctt 28

<210> 21
<211> 622
<212> DNA
<213> unknown

<220>

<223> PCR amplification product using Bacillus subtilis as template
source and SEQ ID NO:18 and SEQ ID NO:20 as primers

<400> 21
gcttgtgact ttgagcgggg cttcattcgt gctgagacag ttgcttatga ggatcttctt 60
gcgggcgggcg gtatggcagg agctaaagag gcaggaaaag tccgccttga agggaaagaa 120
tatgtggtcc aagacggaga tgttattcat ttccgattta atgtatagga tgcagttgta 180
aagggacaag agctttggta taatataaaa ttgtgagtaa tagaattatt gtccttgcc 240
cattatgggc cgcttagtcc aaaaggaggt gcaaacagat gagaaagtac gaagttatgt 300
acattatccg cccaaacatt gacgaagagt ctaaaaaagc agttatcgag cgtttcaaca 360
acgttctgac ttctaacggt gcggagatca ctggaacaaa ggattggggg aaacgctgctc 420
ttgcttacga aatcaacgat ttccgcgacg gtttctacca aatcgtaaac gttcaatctg 480
acgctgcggc agttcaagaa tttgaccgtc tagctaagat cagtgcgat atcattcgcc 540
acattgttgt taaagaagaa gaataagcaa ttttgaaata tataatggta aaaggtggtc 600
tttcttatgc ttaaccgagg at 622

<210> 22
<211> 30
<212> DNA
<213> unknown

<220>
<223> Reverse primer to amplify the rpsF promoter region

<400> 22
gctggatcca tcttcgtcaa tgtttgggcg 30

<210> 23
<211> 331
<212> DNA
<213> unknown

<220>
<223> PCR amplification product using Bacillus subtilis as template
source and SEQ ID NO:20 and SEQ ID NO:22 as primers

<400> 23
gcttgtgact ttgagcgggg cttcattcgt gctgagacag ttgcttatga ggatcttctt 60
gcgggcgggcg gtatggcagg agctaaagag gcaggaaaag tccgccttga agggaaagaa 120
tatgtggtcc aagacggaga tgttattcat ttccgattta atgtatagga tgcagttgta 180
aagggacaag agctttggta taatataaaa ttgtgagtaa tagaattatt gtccttgcc 240
cattatgggc cgcttagtcc aaaaggaggt gcaaacagat gagaaagtac gaagttatgt 300

acattatccg cccaaacatt gacgaagatg g 331

<210> 24

<211> 26

<212> DNA

<213> unknown

<220>

<223> Forward primer to amplify the ywpH promoter region

<400> 24

cccaagcttt caagctgtca atgccg 26

<210> 25

<211> 27

<212> DNA

<213> unknown

<220>

<223> Reverse primer to amplify the ywpH promoter region

<400> 25

cgcgatccg attgaacatg cgattcc 27

<210> 26

<211> 300

<212> DNA

<213> unknown

<220>

<223> PCR amplification product using Bacillus subtilus as template
source and SEQ ID NO:24 and SEQ ID NO:25 as primers

<400> 26

gctttcaagc tgtcaatgcc ggaaaaaaaaa ttgagctttc agtggtttgc gtgggatggc 60

tcttcctatg tgcgcatgaa tacgcaaaac tggtgacaa agcaaattctt tttccgtttt 120

ttaaaaagta catatttctt caaaggaaaa aagcaaaaga tgtttttagc tgaaggaaaa 180

atgaaaacga aagataaaaa cagaggctga aagccatttt taagcgtttt tcttttcttg 240

ttgcatcatt tacaatacat acaaccgcaa ggagaggagg aatcgcatgt tcaatcggat 300

<210> 27

<211> 27

<212> DNA

<213> unknown

<220>

<223> amyE-1: Forward primer

<400> 27

gctctagacg aattatatgg atgtgac 27

<210> 28

<211> 30

<212> DNA

<213> unknown

<220>

<223> amyE-2: Reverse primer

<400> 28

gcctagagct cggatctcct tttccgattg 30

<210> 29

<211> 34

<212> DNA

<213> unknown

<220>

<223> Kn-1: Forward primer

<400> 29

caatctgcag tcgcatgag aatagtgaat ggac 34

<210> 30

<211> 28

<212> DNA

<213> unknown

<220>

<223> Kn-2: Reverse primer

<400> 30

cgggtacctc aaaatggat gcgttttg 28

<210> 31

<211> 269

<212> DNA

<213> unknown

<220>

<223> artificially made spoVG RBS

<400> 31

gaagatctct cgagggtacc ttgatacact aatgctttta tatagggaaa aggtggtgaa 60

ctcatatgaa tcgagcttct agagagctcc catggaacta tgtgattacg aaaatatatc 120

cctttccacc cacttgagta tacttagctc gaagatctct cgagggtacc ttgatacacc 180

taatgctttt atatagggaa aaggtggtgc atggaactat gtgattacga aaatatatcc 240

ctttccacc acttgagtat acttagctc 269

<210> 32
<211> 29
<212> DNA
<213> unknown

<220>
<223> spoVG1 primer

<400> 32
gaagatctct cgagggtacc ttgatacac 29

<210> 33
<211> 30
<212> DNA
<213> unknown

<220>
<223> spoVG2 primer

<400> 33
ctatataaaa gcattagtgt atcaaggtac 30

<210> 34
<211> 30
<212> DNA
<213> unknown

<220>
<223> spoVG3 primer

<400> 34
ctaattgcttt tatataggga aaaggtggtg 30

<210> 35
<211> 30
<212> DNA
<213> unknown

<220>
<223> spoVG4 primer

<400> 35
ctcgattcat atgagttcac caccttttcc 30

<210> 36
<211> 29
<212> DNA
<213> unknown

<220>
<223> spoVG5 primer

<400> 36
 gactgcagct cgaggggtacc ttgatacac 29

<210> 37
 <211> 41
 <212> DNA
 <213> unknown

<220>
 <223> Gntterm-1: Forward primer

<400> 37
 gcggatccag gcctaactaa ttaacctgta ttaaaaacac g 41

<210> 38
 <211> 33
 <212> DNA
 <213> unknown

<220>
 <223> Gntterm-2: Reverse primer

<400> 38
 gctctagagt taaccttctg ttgtttggga tag 33

<210> 39
 <211> 40
 <212> DNA
 <213> unknown

<220>
 <223> B-toxoid3 primer

<400> 39
 aactgcagag atctcatatg aagaaaaaat ttatttcatt 40

<210> 40
 <211> 32
 <212> DNA
 <213> unknown

<220>
 <223> Btoxoid2 primer

<400> 40
 cgcggtacct taaatagctg ttactttgtg ag 32

<210> 41
 <211> 33
 <212> DNA
 <213> unknown

<220>
<223> rpsF-3 primer

<400> 41
ggaattcctg caggtgactt tgagcggggc ttc 33

<210> 42
<211> 24
<212> DNA
<213> unknown

<220>
<223> rpsF-4 and rpsF-5 primer

<400> 42
cgtactttct catatgtttg cacc 24

<210> 43
<211> 28
<212> DNA
<213> unknown

<220>
<223> amyEback-1 primer

<400> 43
cccaagcttt cgacatggat gagcgatg 28

<210> 44
<211> 28
<212> DNA
<213> unknown

<220>
<223> amyEback-2 primer

<400> 44
gcagctcgag gctccggcgc aaatgcag 28

<210> 45
<211> 29
<212> DNA
<213> unknown

<220>
<223> amyEfront-1 primer

<400> 45
cgcgagctca acaaaattct ccagtcttc 29

<210> 46

<211> 31
 <212> DNA
 <213> unknown

 <220>
 <223> amyEfront-2 primer

 <400> 46
 cggctctagaa gtttttaatt tgtgtgtttc c 31

 <210> 47
 <211> 23
 <212> DNA
 <213> unknown

 <220>
 <223> cmR-1 primer

 <400> 47
 cgggaattct catgtttgac agc 23

 <210> 48
 <211> 30
 <212> DNA
 <213> unknown

 <220>
 <223> cmR-2 primer

 <400> 48
 cgccaagctt cccagtagta ggttgaggcc 30

 <210> 49
 <211> 41
 <212> DNA
 <213> unknown

 <220>
 <223> Gntterm-3: Forward primer

 <400> 49
 gcggatccta cgtaaaactaa ttaacctgta ttaaaaacac g 41

 <210> 50
 <211> 315
 <212> DNA
 <213> unknown

 <220>
 <223> upstream region of the rpsF gene, indicating the location of the
 -10 and -35 regions and AT-rich stretches further upstream, as
 well as the RBS and the start codon of the rpsF gene

<400> 50
ctgagtgtgc aggaattatt catagtgact ttgagcgggg cttcattcgt gctgagacag 60

ttgcttatga ggatcttctt gcgggcggcg gtatggcagg agctaaagag gcaggaaaag 120

tccgccttga agggaaagaa tatgtggtcc aagacggaga tggtattcat ttccgattta 180

atgtatagga tgcagttgta aagggacaag agctttggta taatataaaa ttgtgagtaa 240

tagaattatt gtccttgcc cattatgggc cgcttagtcc aaaaggaggt gcaaacagat 300

ggaagttact gacgt 315

<210> 51
<211> 312
<212> DNA
<213> unknown

<220>
<223> upstream region of the ywpH gene

<400> 51
tcaagctgtc aatgccgga aaaaaattga gctttcagtg gtttgcgtgg gatggctctt 60

cctatgtgcg catgaatag caaaactggc tgacaaagca aatctttttc cgttttttta 120

aaagtacata tttcttcaaa ggaaaaaagc aaaagatggt ttagctgaa ggaaaaatga 180

aaacgaaaga taaaaacaga ggctgaaagc catttttaag cgtttttctt ttcttgttgc 240

atcatttaca atacatacaa ccgcaaggag aggaggaatc gcatgttcaa tcaggtcatg 300

cttgtcggac gt 312